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RELATIVE GROWTH RATE AND SURVIVAL, EFFICIENCY OF FEED UTILIZATION OF NILE TILAPIA (*OREOCHROMIS NILOTICUS*), AN EXPERIMENTAL STUDY BY THE ADDITION OF HERBAL EXTRACT SUPPLEMENTS AS DRUGS AND GROWTH PROMOTERS TO JUVENILE FISHES IN THE FED DIET

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ABSTRACT

The use of herbs as drugs in disease management is gaining success, because herbal drugs are cost effective, eco-friendly and have minimal side effects. Many studies have proved that herbal additives enhanced the growth of fishes and protect them from diseases. Indeed, the herbs/herbal drugs overcome all these problems and keep the fish healthy by acting as growth promoters and treating the diseases as well. A feeding trial, involving the use of four different starter diets for the feeding of *Oreochromis niloticus* juveniles, was carried out in the laboratory by intensive cage farming technique. The purpose of this study was to determine the effect of adding herbal extracts in feed and observations were conducted on efficiency of feed utilization (EFU), relative growth rate (RGR), survival rate (SR) of Nile tilapia fish (*Oreochromis niloticus*). Fish samples in this research with an average weight of 5.34 ± 0.16 g were selected. The formulated diets consist of Fresh *Cocos nucifera* milk (CNMe), *Ficus sycomorus* (FSBe), *Suaeda maritima* (SMPe), *Vulva Proliferans* (VPAe), and regular basal diet with all the natural ingredients. The juvenile fishes were distinctly arranged into 6 groups and fed with control, marketed feed also. Every group was fed with formulated basal feed at time intervals of 4hrs thrice a day at 08.00 am, 12.00 pm and 14.00 pm at the ration of around 14.28g/day over a period of 7 days and varied formulated herbal feed for the next 7 days until 84 days study. The results show that the addition of herbal extracts significantly ($P < 0.05$) affected on EFU, RGR on the other hand it insignificantly ($P > 0.05$) affected on SR of Nile tilapia. Based on results, it was concluded that growth performance and feed efficiency increased with the use of *Ficus sycomorus* berries extract and *Suaeda maritima* whole plant extract. *Ficus sycomorus* treatment is highly recommended as live food in the growth promotion of the *Oreochromis niloticus* fish. The proteinaceous food from plants is easily available and most widely cultivable in freshwater fish pond environments, its bio-nutrients are readily available/utilizable by the fish and the procurement is highly economical. This technology uses herbal ingredients in formulation thus contributing towards good quality in fish meal and culturing.

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INTRODUCTION

Products that improve feed efficiency are particularly important since feed costs are a major expense in aquaculture production. Non nutritive feed additives are being used in aquatic feeds to ensure ingestion, digestion, and absorption of dietary nutrients. Feed additives may be both nutritive and non-nutritive ingredients and work by either direct or indirect methods on the animal's system [1-3]. Feed additives are supplemented in small amounts (alone or in combination) for a specific purpose, such as to improve the quality of fish as a final product, to preserve the physical and chemical quality of the diet or to maintain the quality of the aquatic environment.

Nutritional requirements of an animal are a fundamental aspect that depends on species, habitat and live cycle stage [4]. The use of antibiotics and other chemotherapeutics for controlling diseases has been criticized for their negative impacts. Use of herbs as drugs in disease management is gaining success, because herbal drugs are cost effective, ecofriendly and have minimal side effects. Traditional herbal medicines seem to have the potential immune stimulation. Thus, the use of herbs is an alternative to antibiotics in fish health management. Many studies have proved that herbal additives enhanced the growth of fishes and protected them from diseases [5]. The herbs are not only safe for consumers but also widely available throughout Asia and they also have a significant role in aquaculture [6]. The inclusion of herbal feed supplements often provides cooperative action to various physiological functions. The synergistic effect of herbs has been reported in many fishes, including Japanese flounder [7] and *Clarias gariepinus* [8].

Oreochromis niloticus aquaculture needs feed that contains complete nutrients. It should also be efficient and economical it generally requires highly complete feed, which contains fat, protein, carbohydrate, vitamins, and minerals. Protein is the most important element in the feed to support growth. Protein in the feed can be from plant-based protein or animal-based protein. Presently there is no study available on the effects of adding multi nutritional herbal extract diet. Therefore, we evaluated the effect of novel herbal formulated feed on the growth performance and feed utilization of *Oreochromis niloticus* fish.

MATERIALS AND METHODS

The study was conducted at Dept. of Pharmaceutical & Medicinal chemistry Laboratory at Koringa College of pharmacy, Korangi, East Godavari district, India, from August 2018 to October 2018. The completely randomized design was utilized in this study. Five levels of feed dosage of each herbal extract were tested in the concentration range of 20gm, 40gm, 60gm, 80gm, and 100gm/kg of feed along with control. Results of every treatment were recorded. The standard diet was purchased from the local market available in the Koringa village belongs to Kingmei brand and formulated Experimental diet was tested for efficacy **Table-3**.

Management and feeding

Fish used in this study were fingerlings of Nile tilapia (*Oreochromis niloticus*) with the average weight of 5.34 ± 0.16 g. The fish was obtained from Fresh water Canal of Godavari River in the Central zone of Korangi village, Kakinada city. The fish was selected on the base of size uniformity, health, completeness of organs, and lack of potential diseases. The selected fish was placed in to the cultivation container for 7 (seven) days in order to adapt to the environment and fed with normal feed. The density of fingerlings was 3 fish per tank [9]. After 7 (seven) days, the fish was left to fast for one day to empty waste of its metabolism; therefore, at the beginning of the study, the fish could be weighed without any waste of metabolism in the body [10]. The sampled fish was raised for 84 days and fed at satiation 3 (three) times a day at 08:00, 12:00, and 14:00 hrs. The sampled fish was weighed every 7 (seven) days. Containers used in the study were made of virgin food grade plastic with the dimensions of 1 m x 1 m x 0.6 m. The experiment was conducted with 4 (four) treatments, 5 (five) levels. Equipped with aerator to circulate the oxygen in water tank [11]. The study used Biological grade formulated feed in the form of pellet containing protein herbal extracts and added ingredients. Each feed was fed thrice daily (08.00, 12.00, and 16.00h) to approximate satiation for 12 weeks. Initially, fish were fed at 3 times the body weight and subsequently adjusted based on daily intake. Round-the-clock aeration was provided to all the containers from a compressed air pump and manual water exchange was carried out daily.

Water analyses

Water parameters were closely monitored and were kept within safe limits by regular flow management. The water parameters were evaluated in terms of pH, temperature, dissolved oxygen (DO) and salinity. Water was analyzed for its pH by pH metre (systronics pH-207, India), dissolved oxygen (DO) by DO meter (YSI-55, India). Moreover, temperature was recorded by the digital thermometer (H M Digital, Hicks). These observations were recorded twice (morning and noon) daily from different containers.

Feed additives in tilapia nutrition

The feed was made of crude protein meal (sesame oil cake, peanut, soybean), fiber (corn, wheat, ragi flour), lipid (ground nut oil, rice bran oil, palm oil), carbohydrate source (potato starch) and other added vitamins and minerals, Tragacanth as a binder.

Formulation method

The procedure to prepare feed was started with dissolving an appropriate dose of crude herbal extracts into warm water (45°C) and then mixing with additives meal evenly. The mixture was stored in the air-sealed container for 24 h [12]. According to National Research Council (NRC) [13], feed is made by mixing the least amount of the ingredients first and gradually adding and mixing other ingredients with larger amounts except fat sources that are added after all the ingredients have been mixed. The mixture was then formed into granules by passing through a sieve of mesh size 60# with diameter of 1-2 mm. Then, the feed was dried in the oven at 40 °C. The composition and results of proximate analysis of the feed can be seen in Table-1.

Growth and body indices

Growth and morphometric parameters were carefully examined to predict daily feed ration. Each fish was individually weighed and all the other morphometric parameters were measured before the start of the trial and then in the lapse of every week. The fish was removed from the tank individually and placed on laminated graph paper. Length from the mouth to the caudal peduncle and depth from the deepest point of the body to the base of the dorsal fin was measured (mm). Fish were then placed in a tarred beaker of tank water on an analytical balance for body weight measurements and then returned to their respective holding tank. The duration of study was 12 weeks.

Statistical Analysis

Data was subjected to One-way ANOVA Technique under completely randomized design. Graph Pad Prism 5.0 version was used for graphical representation in the experiment. Analysis of variance (ANOVA) was used to analyze the data. Before analysis, the data were first tested for normality, additivity, and homogeneity. The tests were to make sure that the data were normal, homogenous, and had additive property. The analysis of variance was calculated as significant ($p < 0.05$) or highly significant ($p < 0.01$).

RESULTS

Table-1 Proximate Composition of various components available in the Fish meal.

Components	Sesame oil cake	Soyabean meal	Peanut meal	Wheat flour	Corn flour	Milk powder	Vanami Feed
Crude protein	25.7	40.8	20.22	10.62	7.85	35.95	50.12
Crude fiber	10.0	5.30	0.41	0.49	1.96	0.20	25.82
Lipid	10.2	5.71	1.23	1.26	3.82	1.61	15.91
Ash	6.12	6.32	2.49	0.85	1.26	8.43	5.25
Calcium	0.49	0.22	0.04	0.04	0.06	11.9	2.83
phosphorous	0.67	0.56	0.15	0.19	0.29	9.42	3.34
Moisture	8.63	9.35	11.83	12.85	12.05	4.19	4.97

Table-2 Raw materials of formulated herbal diet used in the study calculated for 50gm.

S.no	Ingredients	CNMe	FSBe	SMPe	VPAe	Base meal
1	Corn flour	11g	10g	10g	10g	10g
2	Sesame oil cake	3g	2g	2g	2g	2g
3	Soya bean meal	11g	10g	10g	10g	10g
4	Peanut	3g	2g	2g	2g	2g
5	Wheat flour	6g	5g	5g	5g	5g
6	Starch	4g	3g	3g	3g	3g
7	Milk powder	6g	5g	5g	5g	5g
8	Agar	2g	1g	1g	1g	1g
9	Tragacanth	3g	2g	2g	2g	2g
10	Coconut milk*	L ⁵	----	----	----	----
11	Ficus sycomorus*	----	L ⁵	----	----	----
12	Suaeda maritima *	----	----	L ⁵	----	----
13	Vulva Proliferans*	----	----	----	L ⁵	----
14	Streptomycin	0.3%	0.3%	0.3%	0.3%	0.3%
15	Distilled water	qs	qs	qs	qs	qs
16	Mineral Mix	0.3%	0.3%	0.3%	0.3%	0.3%
17	Vitamin Mix	0.3%	0.3%	0.3%	0.3%	0.3%

Note: *L⁵ Indicates Feed with 5 level extract (Basal feed+ 20, 40, 60, 80,100gm) treatments.

Table-3 Results of the experimental diet tested for efficacy.

Parameter	CNMeL ⁵	FSBeL ⁵	SMPeL ⁵	VPAeL ⁵	Base Meal	Vanami Feed
Body weight (gm)	121.27±0.1	139.19±0.1	128.33±0.1	116.68±0.1	126.14±0.1	142.75±0.1
Length (cm)	20.64±0.1	34.45±0.1	29.34±0.1	27.96±0.1	22.69±0.1	45.12±0.1
EFU	53.09±0.99	58.36±1.03	55.23±0.81	54.71±0.45	53.98±0.96	59.41±0.73
RGR	2.36±0.1	4.75±0.1	4.18±0.1	4.06±0.1	1.86±0.1	5.19±0.1
SR	02	03	03	03	02	03

DISCUSSION

Body weight and body indices of all the groups are shown in Table-3. Significantly growth of the body weight compared with the initial body weight was observed in fish allocated to feed FSB_eL⁵ followed by feed SMP_eL⁵, VPA_eL⁵ and CNM_eL⁵, respectively ($P < 0.05$). All the fish allocated were of equal length and almost weight, mean horizontal growth was not unlike among the various treatments ($P > 0.05$). Caudal length was found to be significantly higher in group fed with berries extract, followed by *Suaeda maritima* whole plant extract, and then *ulva*, coconut milk ($P < 0.05$). More than 10% increase in caudal length has been observed in fish allocated to feed FSB_e as compared to Control basal feed. The girth of fish allocated to group fed with marketed feed vanami was highest while the lowest response was observed in feed with Coconut milk ($P < 0.05$). The addition of *Ficus sycomorus* berries herbal extracts to the basal feed in ascending level of 20,40,60,80,100gm/kg of basal diet significantly ($P < 0.05$) affected increase in girth and caudal length of fish. Similar finding was also reported by the other extracts except with the coconut milk, where addition of copra milk in the feed decreased the weight gain and the relative growth rate was found to be very slow. This copra meal significantly affected on the weight loss. Earlier studies conducted on the effect of dietary medium-and long-chain triacylglycerols (MLCT) on accumulation of body fat in healthy humans by Kasai M *et al* proved that a daily intake of MLCT diet could cause a reduction in body weight and body fat accumulation [14]. In another study on the effect of dietary supplementation with coconut oil on the biochemical and anthropometric profiles of women with abdominal obesity (waist circumferences (WC) > 88cm) the intake of dietary supplement with VCO was observed to decrease the amount of abdominal fat [15].

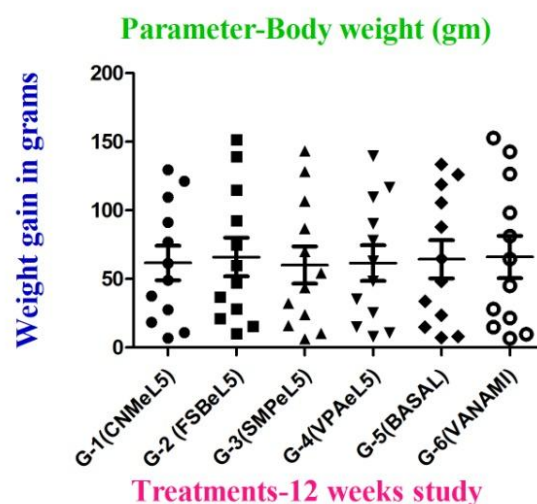


Figure-1: Study of increase in Body weight.

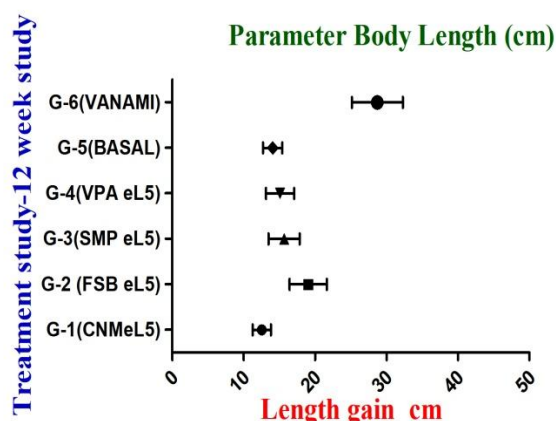


Figure-2: Study of increase in Body length.

The highest efficiency of feed utilization was in treatment FSB_eL⁵ with (100gm/kg feed) with 67.34%, while the lowest was treatment CNMeL⁵ (100gm/kg feed) with 50.15%. treatment with (100gm/kg feed) of Ficus had the highest growth as it was expected, due to the available protein which effectively digest and make available all the nutrients in the meal to build up the body weight. The same results were also reported by Suaeda maritima extract. The high efficiency of feed utilization shows that the fish can efficiently absorb protein to support their growth instead of using it for metabolism, osmoregulation, and reproduction. Growth happens when there is an excess energy from activities for survival, such as breathing, swimming, metabolism, and maintenance. Low efficiency of feed utilization indicated that the fish need more feed to grow. Density and protein content in the feed also influences feed utilization efficiency [16].

$$\text{EFU (\%)} = \frac{\text{Final weight} - \text{Initial weight}}{\text{The amount of feed consumed}} \times 100$$

Parameter-Efficiency of Feed Utilization

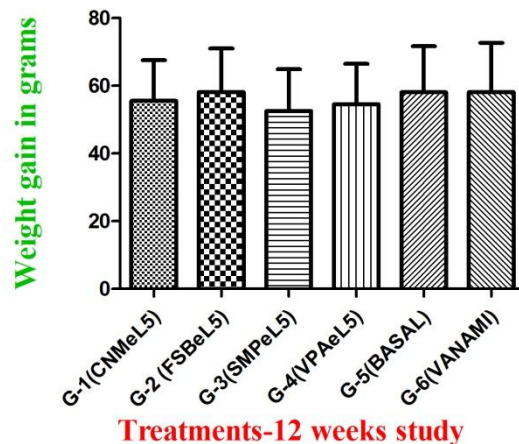


Figure-3: Study of Efficiency in feed utilization.

The Analysis of Variance showed that different doses of extract in the formulated feed had significant effects ($P < 0.05$) on RGR of Nile tilapia (*Oreochromis niloticus*). Growth rate is related to addition of body weight from utilization of protein in the feed. The more protein digested, the higher the energy the fish get to grow [17]. A significant increase in growth was concurrent with increase in extract concentration to a level of 100gm/ kg feed, after which it decreased. It indicated the positive effect on growth performance of the fingerlings in the present study was consistent with the results obtained.

$$\text{RGR (\%)} = \frac{\text{Final weight} - \text{Initial weight}}{\text{Time duration of study}} \times 100$$

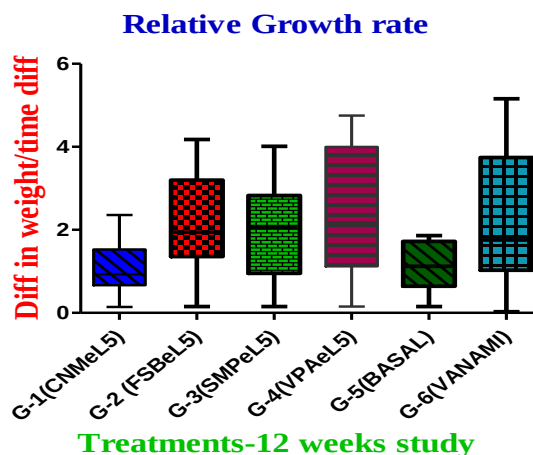


Figure-4: Study of Relative growth rate.

The results of variance analysis showed that addition of herbal extracts in feed insignificantly ($P>0.05$) affected SR of *oreochromis niloticus*. According to Yakuputiya [18], feed is not a factor that influences survival rate, because survival rate is affected by the initial treatment of the fish and quality of the cultivation media. The same results were also reported by Hassan M S *et al.* [19]. Who reported that survival rate was affected by fish gender, heredity, age, reproduction, disease resistance, and external factors such as water quality, density, amount, and composition of amino acids in the feed, quality of Water during the research was on viable condition for tilapia culture. Measurements of water parameter during cultivation are shown in Table-2.

$$SR(\%) = \frac{\text{Final count on fish survived}}{\text{Total fish stocked for study}} \times 100$$

Table 4 Parameters of water quality during the experimental cultivation of *oreochromis niloticus*.

Water Quality			
Treatment	Temperature($^{\circ}$ C)	pH	DO (mg/l)
CNMe	26 – 33	7.50 – 7.85	3.30 – 3.55
FSBe	26 – 33	7.50 – 7.81	3.24-3.48
SMPe	26 – 33	7.50 – 7.82	3.28-3.58
VP Ae	26 – 33	7.50 – 7.87	3.32-3.53
Base meal	26 – 33	7.50 – 7.81	3.26-3.64
Vanami	26 – 33	7.50 – 7.86	3.44-3.84
Feasibility *	14-38*	6.50 – 8.5	>2*

CONCLUSION

As herbs are natural products they are free from side effects, they are comparatively safe, eco-friendly and locally available. Traditionally there are lots of herbs used for various ailments. These herbal products are today the symbol of safety in contrast to the synthetic drugs that are regarded as unsafe to human being and environment. Addition of herbal extracts in feed significantly increased feed utilization, digestibility of feed, and relative growth rate of *Oreochromis niloticus* fish. The optimal doses of extract based on EFU, RGR, in fish culture study were 60, 80, and 100gm/ kg feed respectively.

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